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INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 3 :		(11) International Publication Number: WO 83/ 01119
G01N 35/02, 33/56, 33/54	A1	(43) International Publication Date: 31 March 1983 (31.03.83)

(21) International Application Number: PCT/AU82/00158

(22) International Filing Date: 22 September 1982 (22.09.82)

(31) Priority Application Number: PF 0913

(32) Priority Date: 25 September 1981 (25.09.81)

(33) Priority Country: AU

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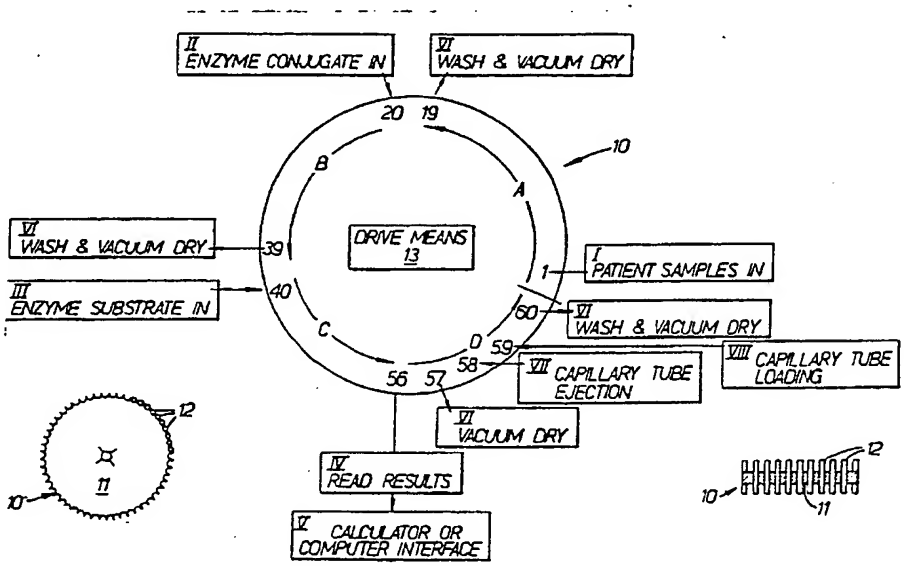
(81) Designated States: AT (European patent), BE (European patent), CH (European patent), DE (European patent), DK, FR (European patent), GB (European patent), JP, LU (European patent), NL (European patent), NO, SE (European patent), US.

Published
With international search report.

(54) Title: AUTOMATED IMMUNOASSAY SYSTEM



(57) Abstract

Apparatus for performing automated heterogeneous immunoassays for the detection or determination of antigenic or haptenic substances or antibodies in a plurality of samples comprising: (a) a plurality of capillary tubes (12), each of the capillary tubes having antibodies or antigenic or haptenic substances attached to the internal surface thereof; (b) means (13) for passing each of the capillary tubes, in sequence, to a plurality of operation stations; (c) means at a first operation station (I) for admitting individual samples, in sequence, to each of the capillary tubes as it is passed to the first operation station; (d) means at one or more subsequent operation stations (II), (III), for admitting immunoassay reagent to each of the capillary tubes as it is passed to the subsequent operation station(s); and (e) means at a final operation station (IV), (V), for detecting or determining the result of the immunoassay in each of the capillary tubes as it is passed to the final operation station. A method of performing immunoassays using this apparatus is also disclosed.

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"AUTOMATED IMMUNOASSAY SYSTEM"

This invention relates to an automated immunoassay system, in particular to a novel immunoassay system which is mechanically simple and compact, and hence relatively inexpensive, and yet is fully automated and capable of handling hundreds of unknown samples per day at a fraction of the cost per test when compared to existing automated systems. Although the system of this invention is suitable for radioimmunoassay (RIA) and other forms of immunoassay, it is particularly suitable for use in enzyme immunoassay by which the disadvantages of RIA including short shelf life of reagents, radioactivity, hazards, waste disposal problems and the like, can be avoided.

At present there are very few fully automated systems for heterogeneous enzyme immunoassays (EIAs), and those that do exist are both cumbersome and expensive. (See, for example, Oellerich M. "Enzyme Immunoassays in Clinical Chemistry: Present Status and Trends", J. Clin, Chem. Clin. Biochem. 1980; 18: 197-208). As a result, at the present time when heterogeneous enzyme

immunoassays are used in hospitals and pathology laboratories, they are most commonly only semi-automated (requiring expensive equipment such as diluters, washers and plate readers), labour intensive and slow. There is therefore an urgent need for a compact, inexpensive, fully automated heterogeneous enzyme immunoassay system capable of handling hundreds of samples per day.

It has now been discovered that a simple and highly effective system can be based on the use of capillary tubes as the solid phase in a heterogeneous enzyme immunoassay system.

According to a first aspect of this invention, there is provided apparatus for performing automated heterogeneous immunoassays for the detection or determination of antigenic or haptenic substances or antibodies in a plurality of samples which comprises:-

(a) a plurality of capillary tubes, each of said capillary tubes having antibodies or antigenic or haptenic substances attached to the internal surface thereof;

(b) means for passing each of said capillary tubes, in sequence, to a plurality of operation stations;

(c) means at a first operation station for admitting individual samples, in sequence, to each of said capillary tubes as it is passed to said first operation station;

(d) means at one or more subsequent operation station(s) for admitting immunoassay reagent to each of said capillary tubes as it is passed to said subsequent operation station(s); and

(e) means at a final operation station for detecting or determining the result of the immunoassay in each of said capillary tubes as it is passed to said final operation station.

In one embodiment of this first aspect of the present invention, there is provided apparatus for performing automated heterogeneous enzyme immunoassays for the detection or determination of antigenic or haptenic substances or antibodies in a plurality of samples which comprises:-

(a) a plurality of capillary tubes, each of said capillary tubes having antibodies or antigenic or haptenic substances attached to the internal surface thereof;

(b) means for passing each of said capillary tubes, in sequence, to a plurality of operation stations;

(c) means at a first operation station for admitting individual samples, in sequence, to each of said capillary tubes as it is passed to said first operation station;

(d) means for admitting enzyme conjugate to each of said samples, said enzyme conjugate being added either to each of said individual samples before said sample is admitted to one of said capillary

tubes, or to each of said individual samples in one of said capillary tubes at a second operation station;

(e) means at a third operation station for admitting enzyme substrate to each of said capillary tubes as it is passed to said third operation station; and

(f) means at a fourth operation station for detecting or determining enzymic action, if any, on said enzyme substrate in each of said capillary tubes.

According to a second aspect of this invention, there is provided a method for performing automated heterogeneous immunoassays for the detection or determination of antigenic or haptenic substances or antibodies in a plurality of samples which comprises:-

(a) providing a plurality of capillary tubes, each of said capillary tubes having antibodies or antigenic or haptenic substances attached to the internal surface thereof;

(b) passing each of said capillary tubes, in sequence, to a plurality of operation stations;

(c) admitting individual samples, in sequence, to each of said capillary tubes at a first operation station;

(d) admitting immunoassay reagent to each of said capillary tubes at one or more subsequent operation stations; and

(e) detecting or determining the result of the immunoassay in each of said capillary tubes at a final operation station.

In one embodiment of this second aspect of the invention, there is provided a method for performing automated heterogeneous enzyme immunoassays for the detection or determination of antigenic or haptenic substances or antibodies in a plurality of samples which comprises:-

(a) providing a plurality of capillary tubes, each of said capillary tubes having antibodies or antigenic or haptenic substances attached to the internal surface thereof;

(b) passing each of said capillary tubes, in sequence, to a plurality of operation stations;

(c) admitting individual samples, in sequence, to each of said capillary tubes at to a first operation station;

(d) admitting enzyme conjugate to each of said samples, either before said sample is admitted to a capillary tube or at a second operation station;

(e) admitting enzyme substrate to each of said capillary tubes at a third operation station; and

(f) detecting or determining enzymic action, if any, on said enzyme substrate in each of said capillary tubes at a fourth operation station.

It will be apparent from the above broad description, that the present invention encompasses assay procedures in which the enzyme conjugate is mixed with the sample before it is added to the capillary tube (for example in the so-called "competitive" or "inhibition" assays - see A(c) below) as well as procedures in which the sample is first added to the capillary tube and enzyme conjugate then added (for example, in the so-called "sandwich" assays - see A(a) and B(a) below). If desired, additional operation stations may also be included to enable the admission of further reagent(s) at appropriate points in the assay procedure. Such further reagents may include, for example, a second antibody where a double antibody type of assay procedure is adopted (see A(b) and B(b) below). In this way, the apparatus and method of the present invention may be adapted to perform a wide variety of assay procedures. The following are illustrative, but by no means limiting, of the types of procedures which may be performed:

A: Antigen detection, e.g. hepatitis, digoxin.

(a) Sandwich antigen assay:

1. Solid phase:	Tube - Anti-hepatitis Ab
2. Specimen:	± Hepatitis subunit or virus
3. Conjugate:	Anti-hepatitis Ab - enzyme

(b) Double antibody sandwich antigen assay:

1. Solid phase:	Tube - Anti-hepatitis Ab Type 1 (e.g sheep antibody)
2. Specimen:	± Hepatitis subunit or virus

3. Second Antibody: Anti-hepatitis Ab Type 2 (e.g. rabbit antibody)
4. Conjugate: Anti-type 2 Ab - enzyme

(c) Competitive antigen assay:
1. Specimen: ± Digoxin
2. Conjugate: Anti-digoxin Ab - enzyme
3. Solid phase: Tube - Digoxin.
(Note: In this assay specimen and conjugate are mixed and incubated prior to addition to tube.)

B: Antibody detection, e.g. tetanus, rubella.

(a) Sandwich antibody assay:
1. Solid phase: Tube - Tetanus Ag
2. Specimen: ± Anti-tetanus Ab (Human)
3. Conjugate: Anti-human Ab - enzyme.

(b) Double antibody sandwich antibody assay:
1. Solid phase: Tube - Tetanus Ag
2. Specimen: ± Tetanus Ab (Human)
3. Second Antibody: Anti-human Ab Type 2 (e.g. sheep antibody against human antibody)
4. Conjugate: Anti-type 2 Ab - enzyme.

Further operation stations may also be interspersed between the first and second operation stations and the second and third operation stations, and at each of these further operation stations may be located means for washing and vacuum drying each of

said capillary tubes before it is passed to the above mentioned second operation station, or third operation station, respectively. Preferably, the various operation stations are suitably located so that appropriate incubation periods are provided as each capillary tube passes from one operation station to the next.

Preferably, the capillary tubes are mounted on a carrier in the form of a spool or carousel having the capillary tubes mounted vertically around the circumference thereof, each capillary tube then forming the reaction vessel (and solid phase) for a single EIA test. In one presently preferred embodiment of such a carrier, the carousel is provided with an appropriate number of outwardly facing slots or detents disposed around its periphery, each slot or detent being dimensioned to receive a capillary tube as a "push-fit" therein, and to releasably retain the tube until it is forced outwardly of the slot. In this way, the capillary tubes may be loaded into the periphery of the carousel, held therein during the immunoassay and later removed to enable fresh tubes to be loaded into the carousel. Suitable loading and unloading means may be provided as described hereinafter. By way of example, a carousel having diameter of approximately 6 cm might be provided with a capacity to hold 60 capillary tubes around the circumference thereof, whilst a carousel of diameter of approximately 40 cm may have a capacity of 240 tubes. Alternatively, however, the carrier may be in

some other configuration, for example, in the form of an elongate, flexible belt somewhat similar to an "ammunition belt" configuration, again having the capillary tubes mounted vertically therein.

In the embodiment of this apparatus in which the carrier is in the form of a carousel, the automated EIA apparatus conveniently includes means for rotating this carousel in a stepwise manner so that each capillary tube at the circumference of the carousel passes, in sequence, the various operation stations which are located around the perimeter of the spool.

The capillary tubes which are used in the apparatus of the present invention may be made of any suitable material such as glass, polyvinyl chloride, polystyrene or other suitable plastics materials. By way of example, the tubes may be 1.5 cm - 2.0 cm in length, and have an internal diameter of about 1mm and an external diameter of about 2mm. Such tubes have a capacity of around 10-15 µl. More generally, however, capillary tubes having a capacity in the range of from 1 to 30 µl may be used in the apparatus of the invention.

Further details of the apparatus of this invention will be apparent from the following description of a preferred embodiment of the invention which is illustrated, by way of example, in the accompanying drawings. In the drawings:-

Figure 1 is a diagramatic representation of a preferred embodiment of the apparatus of the invention indicating the cycle of operations of this embodiment;

Figures 1a and 1b are top and side elevations, respectively (not to scale), of a schematic representation of a carousel device which is preferred for use in the automated apparatus;

Figure 2 is a perspective view of one embodiment of the apparatus of this invention; and

Figure 3 is an enlarged elevational view in section, of a head device for addition of reagent or wash fluid to the capillary tubes together with a vacuum device for drying the tubes.

Referring firstly to Figures 1a and 1b the device 10 comprises a carrier 11 in the form of a carousel having the capacity to hold a plurality of capillary tubes 12 (not all of which are shown) vertically clipped into slots or detents around the circumference of the carousel. As previously mentioned, about 240 such capillary tubes might be held around a spool of diameter of about 40 cm. The internal surface of each of these capillary tubes is coated with antibody or antigenic or haptenic substance by techniques known per se, including adsorption or covalent bonding.

Turning now to Figure 1, there is schematically depicted apparatus in accordance with the preferred embodiment of the present invention

which comprises a device 10 of the type shown in Figures 1a and 1b, and drive means 13 for stepwise rotation of the device 10 so that each of the capillary tubes on the circumference of the device 10 passes, in sequence, the operation stations positioned around the circular path of each capillary tube. Drive means 13 may, for example, comprise a geared motor controlled by a microprocessor to achieve this stepwise rotation. Accurate location of the capillary tubes at each of the operation stations can be effected by means of a detent mechanism operating on unoccupied or unfilled slots or detents in the periphery of the carousel. Further details of each of these operation stations will become apparent from the description of the operation of the apparatus below.

As shown in Figure 2, means including hopper 61 are provided to supply capillary tubes to tube loading station 59 (see Figure 1), the tubes falling from the hopper under gravity and being forced along a guide path and into unoccupied slots or detents on the periphery of the carousel in the vertical plane by a rotating resilient wheel, the vertical position of the tubes in the carousel being controlled by suitably positioned guide plates.

For any given test, it will be readily recognised that the capillary tubes 12 of this device 10 will be coated on the internal surfaces thereof with antibody or antigenic or haptenic substances appropriate for the particular test. Loading of the

unknown samples at the first operation station I at position 1 commences the test procedure. As the apparatus of this invention may be used with any immunoassay procedure, the samples may, for example, be serum, plasma, urine or saliva, or toxins or drugs in solution. Whilst the loading of the tubes may be by any suitable means, it has been found that the use of small sample dispensing cups mounted in a separate cup conveyor belt 62 as shown in Figure 2 enables a plurality of samples to be quickly and readily loaded into the capillary tubes 12. Preferably, each unknown sample to be tested is contained in a sample dispensing cup on the separate conveyor belt 62, each of the sample dispensing cups having a small hole formed in the base thereof. The capillary tube to be loaded is brought into contact with this hole in the base of the cup, for example, by hydraulically lowering the cup under the operation of a cam or microprocessor, at loading bridge 63. The sample which has previously been retained in the cup notwithstanding the small hole in the base thereof, passes by capillary action into the tube. In this way, only a very small volume of each sample is required for testing, and the need for complex sampling and sample loading equipment is avoided. After use, each sample dispensing cup in belt 62 may be removed and replaced by a further cup containing a further unknown sample.

After a sample has been loaded into a first capillary tube, drive means 13 advances the device 10

by one step so that a new capillary tube is brought to the first operation station I and at the same time another sample in a further sample dispensing cup is brought into position at loading bridge 63 for loading into this new capillary tube in the same way as previously described. The loading of subsequent capillary tubes proceeds similarly. It will, of course, be apparent that as the loading of the capillary tubes with samples proceeds, the first capillary tube is being advanced stepwise towards the second operation station II.

In the cycle of operations depicted in Figure 1, before reaching the second operating station II, the first capillary tube reaches an intermediate operation station VI at position 19 at which the tube is positioned over a vacuum outlet. The contents of the tube are evacuated and wash buffer metered through the tube from above. Figure 3 shows in detail a suitable head device 100 for addition of wash fluid to the capillary tubes, as well as a suitable vacuum device 120 for drying the tubes. As shown in this Figure, the device 100 comprises a hydraulically operated piston device 101 mounted in housing 102 and arranged to be moved toward the upper end of capillary tube 12 by hydraulic fluid admitted under pressure to chamber 103 formed by the piston 101 and housing 102 via line 104. Piston 101 is provided with an extension 105 having a concave recess 106 at the extremity thereof within which one end of tube 12 is received as shown. Extension 105 of the piston

extends through an appropriate aperture in the support plate 107 and piston 101 is returned to its starting position under action of return spring 108 on release of the hydraulic pressure in chamber 103. Piston 101 and extension 105 are provided with a fluid passageway 109, extending from recess 106 to inlet line 110, through which appropriate wash fluid may be supplied from a metering pump, (not shown) to capillary tube 12 when the tube is engaged in recess 106. Suitable timing means such as a microprocessor or a mechanical cam device are, of course, provided to supply hydraulic fluid under pressure to chamber 103 and thus move piston 101 into engagement with tube 12, and to then supply wash fluid via passageway 109, before the piston returns to its starting position preparatory to repeating the cycle when another capillary tube 12 is moved into position.

Figure 3 also shows a vacuum device 120 positioned at the lower end of tube 12. This device is similar to head device 100 described above with the exception that the passageway extending to the recess is connected to a vacuum. Timing of the operation of device 120 may be arranged to be prior to, simultaneous with, or slightly behind in time, the operation of device 100. In this manner, tube 12 is emptied and vacuum dried before, simultaneously with or slightly after wash fluid is supplied.

Returning now to Figure 1, after washing and drying, the first capillary tube continues to advance

stepwise and is then passed to the second operation station II at position 20 where it receives a metered quantity of enzyme conjugate, for example from a micropump, an automatic pipette or similar metering device. Preferably this reagent is supplied by means of a head device similar in construction and operation to device 100 previously described. In appropriate situations where a wash station is immediately adjacent a reagent addition station, for example at positions 19, 20 and positions 39, 40 in Figure 1, a single device may be used to simultaneously add wash solution to one capillary tube and to add reagent to the immediately adjacent capillary tube (see devices 65 and 66 in Figure 2).

The first capillary tube then advances to a further intermediate operation station VI at position 39 where the capillary tube is again vacuum dried and washed in the same manner as previously described. The tube then advances to the third operation station III at position 40 at which a metered quantity of enzyme substrate is added to the capillary tube. Once again, this may, for example, be effected by means of a micropump, an automatic pipette or similar metering device operating through a head device as previously described.

The tube again advances to the fourth operation station IV at position 56 at which the result of the test which has been performed in that tube is read, for example by a colorimetric reading

taken either through or along the length of the capillary tube. Such a reading may, for example, be accomplished by the use of chopped light from a suitable lamp focused through the capillary tube to impinge on a photodetector. The readout from the colorimeter is passed to a calculator or computer interface V, before being displayed at display panel 64 (Figure 2) or recorded by suitable print-out means.

Suitable switching or other actuating means will, of course, be provided to ensure that the metering or other device at each operating station is actuated to operate on each capillary tube as it arrives at that station.

From the above description, it will be appreciated that each capillary tube of the device 10 passes through three incubation phases, the first incubation phase being indicated in Figure 1 by the letter A and extending from positions 2 to 18; the second incubation period being indicated by the letter B and extending from positions 21 to 38; and the third incubation period being indicated by the letter C and extending from positions 41 to 55. In addition, as each of the capillary tubes advances stepwise, it passes through the various operation stations and the incubation periods in sequence.

If desired, when commencing the cycle of operations with a given device 10, the operation of each of the operation stations may be arranged to

commence as the first capillary tube is advanced from station to station by means of a microswitch positioned at each of the operation stations. The advancing capillaries would activate each operation, for example by activating the circuit of a solenoid-operated micropump for the particular reagent addition required. The passage of the final capillary tube of the device 10 would allow the inactivation of each operation after that final capillary tube has passed the particular operation station.

On completion of the reading of the test in each tube, the tube passes to station VI where it is vacuum dried before passing to tube ejection station VII where it is forced outwardly from the slot or detent in which it was held, for example by means of an unloading device angled to the periphery of the carousel and positioned to engage each capillary tube in turn and force it outwardly.

In a modified embodiment, not shown, the device 10 could be replaced by a toothed wheel of similar size around which would be fed capillary tubes in an "ammunition belt" configuration, the capillary tubes and belt being discarded after use. It will, of course, be appreciated that the same advantages arise from the use of the present invention in this modified embodiment.

The cycle of operation which is described above with reference to Figure 1 of the drawings

illustrates the sequence of steps which are used with assays of the type used for quantitation of rubella and tetanus immunity. For each of these assays, periods of approximately ten minutes are used for the incubation periods, A, B and C, so that, after completing the first cycle of 30 minutes, tests may be completed at the rate of about 120 per hour using a device 10 having 60 capillary tubes mounted thereon. If the diameter of the spool is increased to around 30cm, the throughput can be increased to around 600 tests per hour. Of course, by suitably arranging the positions of the various operation stations and the speed at which the capillary tubes are passed to each operation station, the various incubation periods which are required for other types of assays could be accommodated. Overall, however, the use of capillary tubes in accordance with the present invention has a very significant advantage in that it enables very short incubation periods to be utilised because of the large reactive surface area and small reaction volume of the tube. The short incubation periods therefore make possible a large throughput of tests utilising the apparatus of the present invention.

A further and most significant advantage of the use of capillary tubes in accordance with the present invention arises from the fact that the capillaries require only very small quantities of the reagents, (for example in the range of 1 to 30 µl, particularly of the order of 10 to 15 µl) and so the amounts of reagents which are required to conduct a

series of tests in this apparatus are minimised. The capillary tubes, of course, form open-ended reaction vessels and will automatically pick up and hold their own volume of reagent which may later be emptied simply by passing the tube over a vacuum outlet. As a result, the equipment for loading and washing the tubes may be simple, compact and inexpensive. In contrast, in existing automated EIA systems, reagents in the 100 to 200 µl quantities are metered and dispensed into tubes which, after incubations ranging from 30 minutes upward in a heated water bath, must be evacuated by retractable needles inserted into the tubes. Equipment for these tests is therefore complex, cumbersome and expensive.

It will be appreciated that variations to the sequence of operating stations illustrated in Figure 1 may be made as desired in order to accommodate alternative assay procedures. Thus, in a "competitive" or "inhibition" type of assay, the operation station at which the enzyme conjugate is added to the individual samples is relocated so as to operate at a point on the cup conveyor belt or turntable prior in sequence to addition of the samples to the capillary tubes. In this modification, the conjugate can be added to the samples and incubated prior to addition of the individual mixtures to the tubes. Similarly, additional operation stations may be included at appropriate positions in the cycle of operation for addition of a second antibody in the "double antibody" type of assay.

In one particularly preferred embodiment of the apparatus of this invention, the various operation stations at which the steps of the method are performed are so constructed that the locations of the individual operation stations may be adjusted as desired to different points either along the sample path (for example, where enzyme conjugate is to be mixed and incubated with the samples prior to addition to the capillary tubes) or along the path of the capillary tubes themselves (for example, where the samples are added to the capillary tubes prior to addition of enzyme conjugate thereto). Such adjustment enables appropriate incubation times to be readily and easily selected for the various steps of the test sequence. The adjustment of location of the various operation stations may, for example, be achieved by providing a continuous rod or other support alongside the sample path and/or the path of the capillary tubes, and by so constructing the various operation stations that they can be located at any desired position along this support.

The following examples illustrate the use of the apparatus and method of the present invention in heterogeneous enzyme immunoassays:

EXAMPLE 1 Estimation of Tetanus Immunity in blood from human patients.

This estimation is performed in apparatus of the type illustrated in the accompanying drawings. The

capillary tubes are of 1.5cm length and internal diameter 1.00mm, having a capacity of about 10-15µl. The following test system is used:

Solid phase:	Tube - Tetanus Ag
Specimen:	± Anti-tetanus Ab (IgG)
Conjugate:	Anti-human IgG - urease
Substrate/Indicator:	Urea solution containing Bromocresol purple.

The specimens (human blood) were diluted 1:2 in anticoagulant buffer prior to testing. The cycle of operations is as shown in Figure 1, and the tests performed at room temperature with a 10 minute incubation period between addition of specimens to the tubes and washing the vacuum drying, and again between addition of conjugate and washing and drying, and between addition of substrate/indicator and reading of results.

The results obtained give good correlation when ranked against known standards.

EXAMPLE 2 Estimation of Digoxin in human serum.

This estimation is performed using the same capillary tubes as described above, but in a "competitive" type assay in which the cycle of operations illustrated in Figure 1 is modified so that the enzyme conjugate is added to the specimen and incubated prior to addition of the resulting mixtures

to the tubes. Again, 10 minute incubation periods at room temperature are used throughout the tests. The following test system is used:

Specimen:	± Digoxin
Conjugate:	Anti-digoxin Ab- urease
Solid Phase:	Tube - Digoxin
Substrate/Indicator:	Urea solution containing Bromocresol purple.

This system has been found to give a very clear detection of digoxin in the 0.64 - 6.4 n moles/litre range in human serum.

Other enzymes may of course be used in place of urease as described above, and a number of marker enzymes for use in enzyme immunoassays have previously been described (see Oellerich: Enzyme Immunoassays in Clinical Chemistry, supra.). The use of enzymes such as urease, horse radish peroxidase, alkaline phosphatase and β-galactosidase is, however, preferred as these enzymes can be determined using indicators which provide results which can be read spectrophotometrically. It will be understood, however, that other enzymes and other detection methods, e.g. fluorimetry, luminescence measurement, turbidimetry, potentiometry and thermometry, may be adopted if desired.

It will also be understood that whilst the present invention relates principally to apparatus and methods for performing heterogeneous enzyme

immunoassays characterised by the use of a plurality of capillary tubes, the use of such a plurality of capillary tubes may also be applied to other immunoassays such as radioimmunoassays and homogeneous enzyme immunoassays, and to other general clinical chemistry procedures. In homogeneous enzyme immunoassays, of course, no antibody or antigenic substance coating on the internal surfaces of the tubes is required. For both homogeneous EIA and general clinical chemical assays, the tests are performed in accordance with known techniques, preferably in a series of cups located in a cup conveyor belt or on a turntable, and the final reaction systems then transferred to the capillary tubes which simply become cuvettes in which the enzyme activity or other reaction result is read or estimated by spectrophotometry or one of the other known techniques.

Those skilled in the art will appreciate that the invention described herein is susceptible to other variations and modifications other than those specifically described without departing from the broad teaching herein. It is to be understood that the invention includes all such variations and modifications which fall within its spirit and scope.

CLAIMS:

1. Apparatus for performing automated heterogeneous immunoassays for the detection or determination of antigenic or haptenic substances or antibodies in a plurality of samples which comprises:-

(a) a plurality of capillary tubes, each of said capillary tubes having antibodies or antigenic or haptenic substances attached to the internal surface thereof;

(b) means for passing each of said capillary tubes, in sequence, to a plurality of operation stations;

(c) means at a first operation station for admitting individual samples, in sequence, to each of said capillary tubes as it is passed to said first operation station;

(d) means at one or more subsequent operation station(s) for admitting immunoassay reagent to each of said capillary tubes as it is passed to said subsequent operation station(s); and

(e) means at a final operation station for detecting or determining the result of the immunoassay in each of said capillary tubes as it is passed to said final operation station.

2. Apparatus as claimed in claim 1, wherein said capillary tubes are mounted on a carrier, said

carrier comprising a spool or carousel having said capillary tubes mounted vertically around the periphery thereof.

3. Apparatus as claimed in claim 2, wherein said means for passing each of said capillary tubes to a plurality of operations comprises means for rotating said spool or carousel in a stepwise manner.

4. Apparatus as claimed in claim 2 or claim 3, wherein each of said capillary tubes is releasably retained by frictional engagement in an outwardly directed slot or detent at the periphery of said spool or carousel.

5. Apparatus as claimed in claim 4, further comprising means for loading individual capillary tubes into individual slots or detents of said spool or carousel prior to passage of said tubes to said operation stations, and means for unloading said capillary tubes from said spool or carousel on completion of the immunoassay.

6. Apparatus as claimed in any one of claims 1 to 5 further comprising means to supply said plurality of samples, said means comprising a plurality of sample dispensing cups mounted in cup conveyor means and drive means to advance each of said sample dispensing cups in turn to said first operation station, each of said sample dispensing cups having a small aperture in the base thereof and the sample

being retained therein by capillary action, the apparatus further comprising loading means at said first operation station to contact each capillary tube in turn with the base of a respective one of said dispensing cups so that the sample in said dispensing cup is loaded into said capillary tube.

7. Apparatus as claimed in any one of claims 1 to 6, further comprising washing and/or vacuum drying means positioned at selected points along the pathway of the capillary tubes intermediate said operation stations.

8. Apparatus as claimed in claim 7, wherein each of said washing and/or vacuum drying means includes a piston device movable within a housing to contact each of said capillary tubes in turn as it is passed to said washing and/or vacuum drying means to admit washing fluid to said tube and/or to remove washing and test fluids from said tube.

9. Apparatus as claimed in any one of claims 1 to 8, wherein each of said capillary tubes has a capacity of 1 - 30 µl, preferably 10 - 15 µl.

10. Apparatus for performing automated heterogeneous enzyme immunoassays for the detection or determination of antigenic or haptenic substances or antibodies in a plurality of samples which comprises:-

(a) a plurality of capillary tubes, each of said capillary tubes having antibodies or antigenic or haptenic substances attached to the internal surface thereof;

(b) means for passing each of said capillary tubes, in sequence, to a plurality of operation stations;

(c) means at a first operation station for admitting individual samples, in sequence, to each of said capillary tubes as it is passed to said first operation station;

(d) means for admitting enzyme conjugate to each of said samples, said enzyme conjugate being added either to each of said individual samples before said sample is admitted to one of said capillary tubes, or to each of said individual samples in one of said capillary tubes at a second operation station;

(e) means at a third operation station for admitting enzyme substrate to each of said capillary tubes as it is passed to said third operation station; and

(f) means at a fourth operation station for detecting or determining enzymic action, if any, on said enzyme substrate in each of said capillary tubes.

11. A method for performing automated heterogeneous immunoassays for the detection or determination of antigenic or haptenic substances or antibodies in a plurality of samples which comprises:-

(a) providing a plurality of capillary tubes, each of said capillary tubes having antibodies or antigenic or haptenic substances attached to the internal surface thereof;

(b) passing each of said capillary tubes, in sequence, to a plurality of operation stations;

(c) admitting individual samples, in sequence, to each of said capillary tubes at a first operation station;

(d) admitting immunoassay reagent to each of said capillary tubes at one or more subsequent operation stations; and

(e) detecting or determining the result of the immunoassay in each of said capillary tubes at a final operation station.

12. A method for performing automated heterogeneous enzyme immunoassays for the detection or determination of antigenic or haptenic substances or antibodies in a plurality of samples which comprises:-

(a) providing a plurality of capillary tubes, each of said capillary tubes having antibodies or antigenic or haptenic substances attached to the internal surface thereof;

(b) passing each of said capillary tubes, in sequence, to a plurality of operation stations;

(c) admitting individual samples, in sequence, to each of said capillary tubes at a first operation station;

(d) admitting enzyme conjugate to each of said samples, either before said sample is admitted to

a capillary tube or at a second operation station;

(e) admitting enzyme substrate to each of said capillary tubes at a third operation station; and

(f) detecting or determining enzymic action, if any, on said enzyme substrate in each of said capillary tubes at a fourth operation station.

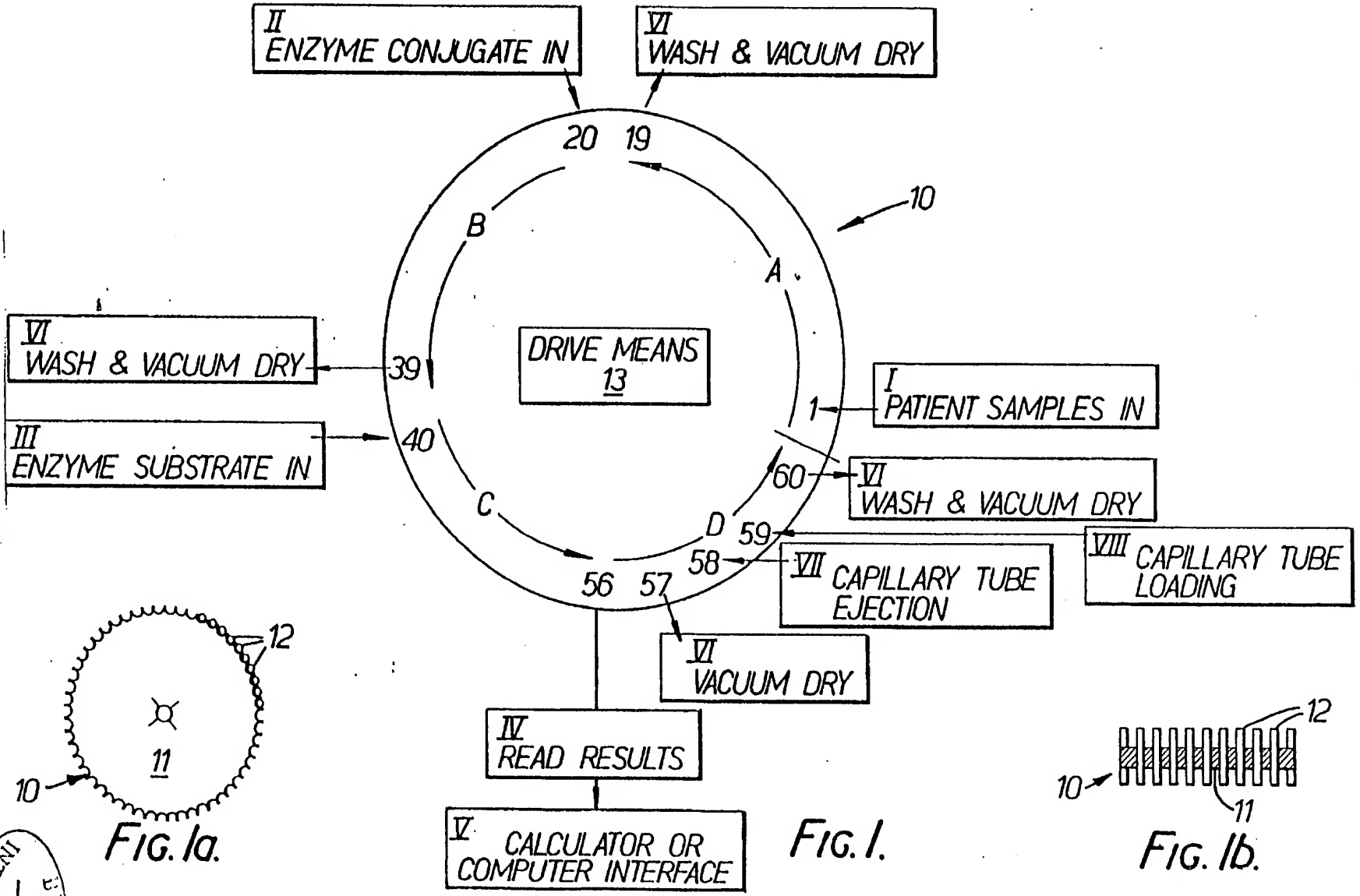

Fig. 1.

Fig. 1a.

Fig. 1b.

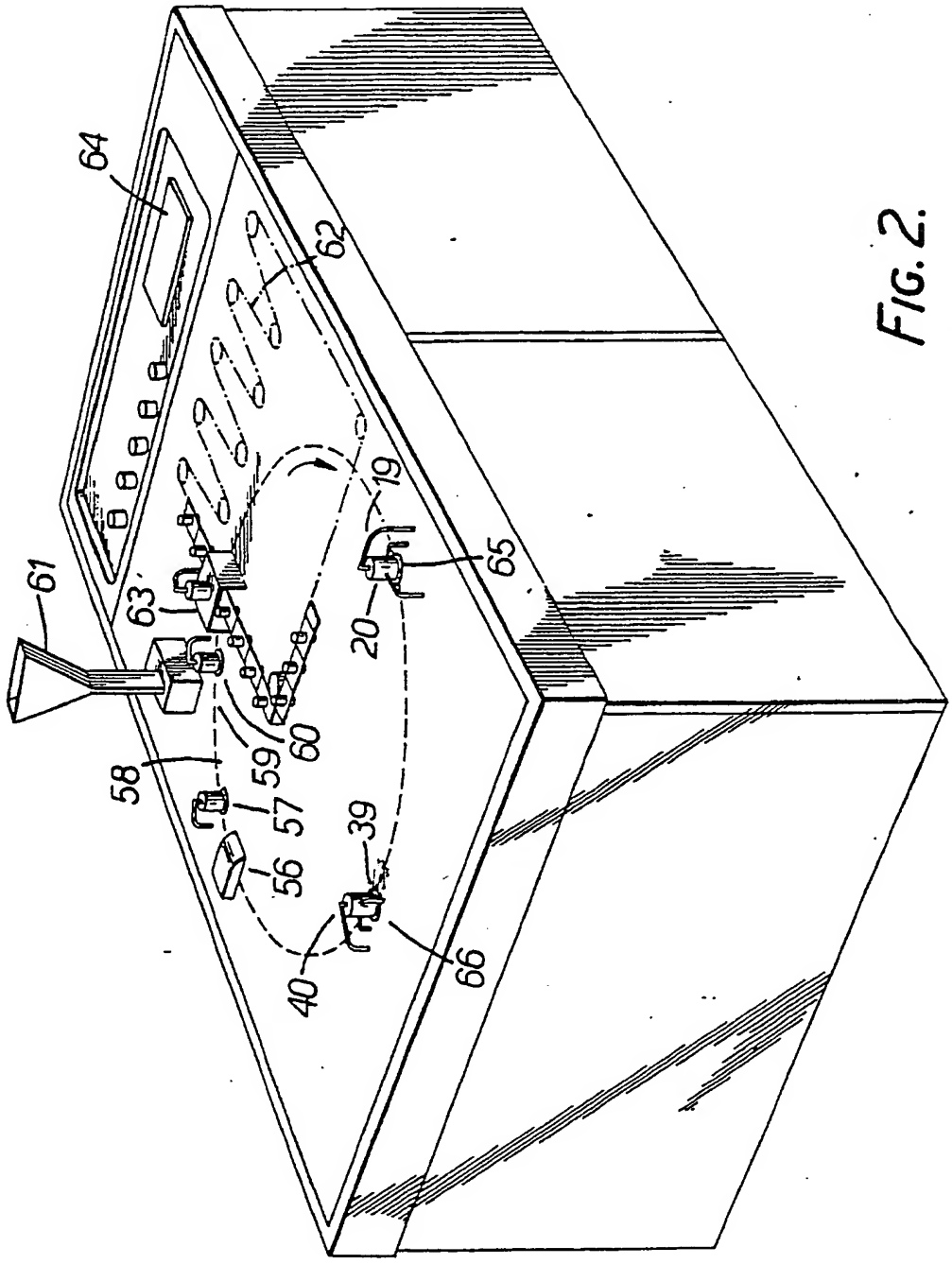

FIG. 2.

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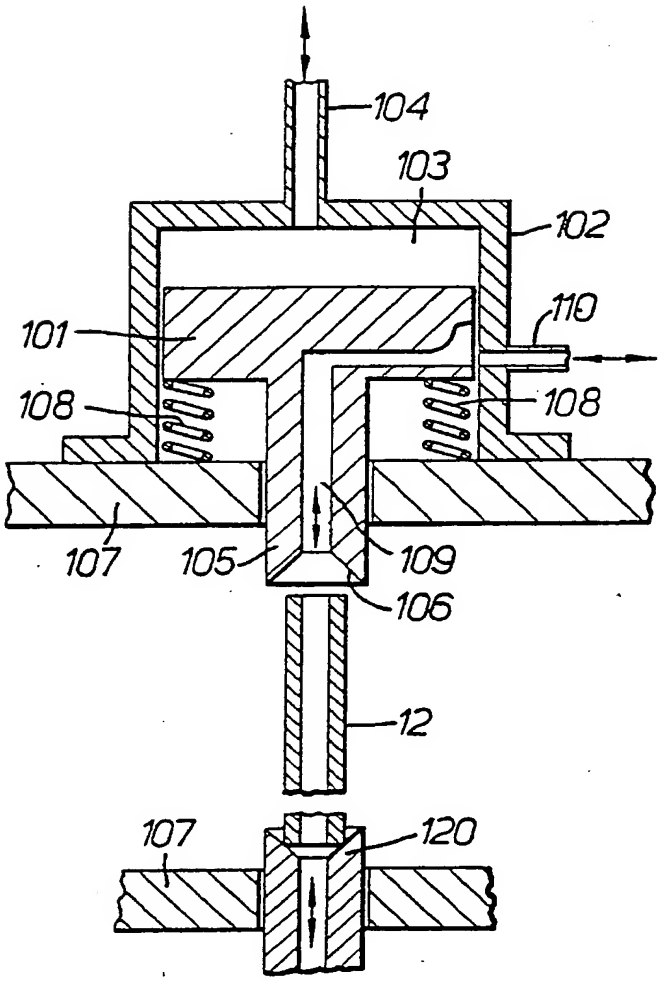

FIG. 3.

INTERNATIONAL SEARCH REPORT

International Application No PCT/AU82/00158

I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all) [3]

According to International Patent Classification (IPC) or to both National Classification and IPC

IPC[3] G01N 35/02, 33/56, 33/54

II. FIELDS SEARCHED

Minimum Documentation Searched [4]

Classification System	Classification Symbols
IPC[3] 2,1	G01N 35/02, 33/56, 33/54
IPC 2,1	G01N 33/16, 31/14
US CL	435.7

Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched [5]

AU:IPC as above

III. DOCUMENTS CONSIDERED TO BE RELEVANT [14]

Category *	Citation of Document, [16] with indication, where appropriate, of the relevant passages [17]	Relevant to Claim No. [18]
P,Y	AU,A, 78678/81 (COMMONWEALTH SERUM LABORATORIES COMMISSION) 1 July 1982, (01.07.82)(& WO,A, 8202211).	(1-12)
Y	CLINICAL CHEMISTRY, VOLUME 25,No7, issued 1979 (Amer Assoc, Clin-Chem,Winston-Salem,N.C.) D. Wagner, B. Alspector, J. Feingers, A.Pick, "Direct Radio-immunoassay with Capillary Chromatography Tubes", see pages 1337 to 1339.	(1)
A	Biological Abstracts, Volume 69, No. 7, issued 1980 (Bio Sciences Information Service, Philadelphia, Penn USA) A.E. Buhl, L.M. Pasztor, J.A. Resko, "Sex steroids in guinea pig fetuses after sexual differentiation of the gonads", see the Abstract No.42629, BIOL REPROD 21/4 : 905-908. 1979.	(1)
A	FR,A 1, 2389134 (SEROA) 24 November 1978 (24.11.78)	(1-12)
A	AU,B, 49119/72 (467849)(UNITED STATES ATOMIC ENERGY COMMISSION) 23 May 1974 (23.05.74).	(1-12)
Y	US,A, 3859051 (ROHE SCIENTIFIC CORP) 7 January 1975 (07.01.75)(and US,A, 3915652)	(1-10)

* Special categories of cited documents: [15]

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

IV. CERTIFICATION

Date of the Actual Completion of the International Search [1]	Date of Mailing of this International Search Report [2]
3 December1982 (03.12.82)	23 December 1982 (23.12.82)
International Searching Authority [1]	**Signature of Authorized Officer [20]**
AUSTRALIAN PATENT OFFICE	P.F. GOTHAM

Form PCT/ISA/210 (second sheet) (October 1981)

FURTHER INFORMATION CONTINUED FROM THE SECOND SHEET

Y	AU,B, 58817/80 (516431) (AMES-YISSUM LTD) 12 March 1981 (12.03.81)	(1)
A	FR,A1, 2397636 (GIST-BROCADES N.V.) 9 February 1979 (& DE,A, 2831083, & JP,A, 54048592 & GB,A, 2005831, & AU,A, 38041/78)	(1-12)

V. ☐ OBSERVATIONS WHERE CERTAIN CLAIMS WERE FOUND UNSEARCHABLE [10]

This International search report has not been established in respect of certain claims under Article 17(2) (a) for the following reasons:

1. ☐ Claim numbers _____, because they relate to subject matter [12] not required to be searched by this Authority, namely:

2. ☐ Claim numbers _____, because they relate to parts of the International application that do not comply with the prescribed requirements to such an extent that no meaningful International search can be carried out [13], specifically:

VI. ☐ OBSERVATIONS WHERE UNITY OF INVENTION IS LACKING [11]

This International Searching Authority found multiple inventions in this International application as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this International search report covers all searchable claims of the International application.

2. ☐ As only some of the required additional search fees were timely paid by the applicant, this International search report covers only those claims of the International application for which fees were paid, specifically claims:

3. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International search report is restricted to the invention first mentioned in the claims; it is covered by claim numbers:

4. ☐ As all searchable claims could be searched without effort justifying an additional fee, the International Searching Authority did not invite payment of any additional fee.

Remark on Protest

☐ The additional search fees were accompanied by applicant's protest.

☐ No protest accompanied the payment of additional search fees.